

WHAT IS CLAIMED IS:

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1. A method to prepare isolated mammalian epidermal stem cells, comprising:
(a) separating in a sample comprising a population of mammalian epidermal cells, a population comprising epidermal stem cells from at least one population of cells that does not comprise epidermal stem cells; and
(b) isolating a substantially pure population of epidermal stem cells from the population of epidermal stem cells.

2. A method to prepare isolated mammalian epidermal stem cells, comprising:
(a) separating in a sample comprising a population of mammalian epidermal cells, a population in the sample which represents the smallest 30% of the cells in the sample and which population comprises epidermal stem cells, from larger cells in the sample; and
(b) isolating a substantially pure population of epidermal stem cells from the smaller cells.

3. The method of claim 1 or 2 wherein the mammalian cells are murine cells.
4. The method of claim 1 or 2 wherein the mammalian cells are human cells.
5. The method of claim 1 or 2 wherein the mammalian cells are primate cells.

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6. The method of claim 1 or 2 wherein the population of mammalian epidermal cells is dissociated from non-epidermal cells prior to separation.
7. The method of claim 1 or 2 wherein the population of mammalian epidermal cells is contacted with Hoechst dye prior to separation.

8. The method of claim 7 wherein the dye is Hoechst 33342.

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9. The method of claim 1 or 2 wherein the population of mammalian epidermal cells is contacted with propidium iodide prior to separation.

10. The method of claim 7 wherein the cells are further contacted with propidium iodide prior to separation.

11. The method of claim 1 or 2 wherein the separation is performed with a flow cytometer.

12. The method of claim 1 or 2 wherein the population of mammalian epidermal cells is in a medium which lacks azide.

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13. The method of claim 1 or 2 wherein the population of mammalian epidermal cells is contacted with a nuclear-retained label prior to separation.


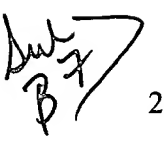
14. The method of claim 1 or 2 wherein the cells that are not epidermal stem cells have proliferative capacity.

15. Epidermal stem cells isolated by the method of claim 1 or 2.

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16. A method to prepare isolated mammalian epidermal stem cells, comprising:
(a) contacting a population of mammalian epidermal cells comprising epidermal stem cells with an amount of a first agent under conditions effective for viable cells to retain the first agent;
(b) contacting the population of (a) with an amount of a second agent under conditions effective for non-viable cells to retain the second agent; and
(c) separating the population of (b) into a population of viable epidermal stem cells and at least one population of cells that does not comprise epidermal stem cells.

17. The method of claim 16 further comprising isolating the epidermal stem cells.

18. The method of claim 16 wherein the mammalian cells are murine cells.
19. The method of claim 16 wherein the mammalian cells are human cells.
20. The method of claim 16 wherein the mammalian cells are primate cells.
21. *Sub B6*  The method of claim 16 wherein the population of mammalian epidermal cells is dissociated from non-epidermal cells prior to step (a).
22. The method of claim 16 wherein the population of mammalian epidermal cells of step (a) is contacted with Hoechst dye.
23. The method of claim 22 wherein the dye is Hoechst 33342.
24. The method of claim 16 wherein the second agent is propidium iodide.
25. *Sub B7*  The method of claim 16 wherein the population of mammalian epidermal cells is contacted with a nuclear-retained label prior to step (a).
26. The method of claim 16 wherein the cells that are not epidermal cells have proliferative capacity.
27. Isolated epidermal stem cells obtained by the method of claim 16.
28. A method of using isolated epidermal stem cells, comprising:
(a) contacting the isolated stem cells of claim 15 or 27 with an isolated nucleic acid molecule comprising an open reading frame so as to yield transformed epidermal stem cells; and
(b) identifying transformed epidermal stem cells.

29. The method of claim 28 wherein the isolated stem cells are contacted with a recombinant virus comprising the nucleic acid molecule.
30. The method of claim 26 wherein the nucleic acid molecule comprises a marker gene.
31. The method of claim 28 wherein the nucleic acid molecule comprises a therapeutic gene.
32. Transformed epidermal stem cells prepared by the method of claim 16.
33. A method to prepare a tissue *in vitro*, comprising:
contacting the epidermal stem cells of claim 15, 27 or 32 with a substrate so as to yield a tissue.
34. The method of claim 33 wherein the substrate comprises fibroblasts.
35. The method of claim 33 wherein the substrate is connective tissue.
36. Tissue produced by the method of claim 33.
37. A method of expressing an open reading frame in a mammal, comprising:
(a) contacting the mammal with the transformed epidermal stem cells of claim 32; and
(b) detecting or determining whether the mammal expresses the open reading frame.
38. A method to prepare a chimeric non-human mammal, comprising:
(a) introducing the epidermal stem cells of claim 15, 27 or 32 into a non-human mammalian blastocyst to form a chimeric blastocyst; and

(b) introducing the chimeric blastocyst into a female non-human mammal capable of gestating a blastocyst to term so as to yield a progeny chimeric mammal.